Caroline Ajo-Franklin Lab Manual Version 1.0

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1. Welcome to the Ajo-Franklin Group!

A. Guiding Principles:

The Ajo-Franklin group is a very collaborative and congenial group.

Your core responsibilities:

- 1. Keep yourself & your colleagues safe in the lab.
- 2. Treat your labmates with respect and collegiality.
- 3. Help people when you can.
- 4. Be productive. Essential elements of this are:
 - To think about what you are doing (constantly) and ask questions whenever you are unsure of why or what you're doing,
 - To keep an organized lab notebook containing observations as well as a written record of what you've, and
 - Strive to work enough hours that you're maximizing your productivity and not so many that it's counterproductive.
- 5. Contribute to exciting new research and discovery!

What you can expect in return:

- 1. To be safe in the lab.
- 2. To be treated with respect and collegiality.
- 3. To be mentored and trained so that you can hone your ability to do scientific research and to communicate about scientific research.
- 4. Contribute to exciting new research and discovery!

2. Emergency Contacts for Current Personnel

Emergency Lab Phone	7-911			Н	ealth Se	rvice	S	6266	
Emergency Cell Phone	911			Ε	Enviro. Health &		&	5514	
				S	Safety				
				S	ecurity			4855	
Lab Location	5204	5208	5210		5216 /5218 5220 Cold Rm			n	
Lab Phone Number	495.2287	495.229	1 495.229	94	none		495.229	08	
		Prefer	red phone a	# ar	nd email	are b o	old		
Staff	Office	Office	Home		Cell LBL Email				
Delegate at least at a	Location	ext.	Phone	P	hone	Oth	er Email		
Principal Investigator	07.5445	400	T 540	1 5	40.000	0.4:	- F		
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NavaDia Lah Managar				<u> </u>					
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Titta Gardia	07.5111	4125			514	IXEC	ar orace in	31.gov	
NanoBio Prin. Scientific									
Engineering Associate									
Michael Connolly	67:5112	486- 6388				mdo	connolly@	<u>ଆbl.gov</u>	
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Postdoctoral Fellows									
Behzad Rad	67:5112	486-			18.422. 241	brac	d@lbl.gov	<u>v</u>	
Graduate Students		T	T			1			
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Hardones de como de co	Out a Arr	Tables		<u> </u>					
Undergraduate Students	Cube Area			0	OF 040	nha	adla@ba	rkolov odu	
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Sean Stettner	67:5108	486- 6907			70-290- 755	sste	ttner@lbl.	gov	
Other Associated Personnel	ated								
Ramya Tunuguntla	67:5108 C	486- 6907							
Karina Sand									

3. Contact Information for Lab Alumni

Lab Alumni are part of our extended family. Please see the Ajo-Franklin website:

http://www.ajofranklin.com/caroline/CAjoFranklinLab.html

4. Lab Policies

A. Your core responsibilities:

- Keep yourself & your colleagues safe in the lab.
- Treat your labmates with respect and collegiality.
- Be productive. Essential elements of this are:
 - To think about what you are doing (constantly).
 - Ask questions whenever you are unsure of why or what you're doing.
 - To keep an organized lab notebook containing observations as well as a written record of what you've done, are doing and plan to do.
 - Strive to work enough hours that you're maximizing your productivity and not so many that it's counterproductive.
- Contribute to exciting new research and discovery!

What you can expect in return:

- To be safe in the lab.
- To be treated with respect and collegiality.
- To be mentored and trained so that you can hone your ability to do scientific research and to communicate about scientific research.
- Contribute to exciting new research and discovery!

B. <u>Common Sense Rules</u> (reference: At the Bench: Chapter 1. Survival through Common Sense and Courtesy)

- Everyone in the lab is willing to help, but extremely busy. Please respect this and plan accordingly.
- E-mail requests as well as asking verbally.
- Ask, don't command. Treat staff and students as co-workers.
- Assume nothing. Don't assume someone will stop their work to help you whenever you need it. Be humble in your expectations. Also don't assume everyone else is always right.
- Plan ahead! Make an appointment with lab staff & equipment managers in advance for training.
 - o E-mail requests as well as verbal

- o They will need to reserve time on their busy calendars and the equipment calendar.
- o Some training is best with real samples, ask your equipment manager.
- o know the potential hazards of the chemicals, biologicals or processes you will be using
- Write down detailed notes when getting instruction! Avoids unnecessary questions later.
 - ✓ Names, contact info, locations
 - ✓ Name of instrument and model (great for paper writing, and manual look up)
 - ✓ Times, temperatures, reagent details and locations
 - ✓ Diagrams of set-ups
 - ✓ This also shows the goodwill that you are interested in the instruction!
- If you don't know how to use something, forget, or are unsure about a chemical, its waste disposal, or procedure, ask for help!
- Do not discuss lab member's results or proposals with others not in the lab without permission. No one
 wants to get scooped before publishing or they may need to verify and repeat experiments.

C. Equipment Training

- If you don't know how to use a piece of equipment, no matter how small, ask for training! Then take detailed notes, diagrams, location, contacts, everything etc.....
 - This avoids so problems with:
 - Safety
 - Equipment damage
 - Having to re-do experiments
- Many pieces of equipment in the laboratory REQUIRE training before use, and calendar reservations.
 - o We use resource data base for reserving instruments
 - https://resourcedb.lbl.gov/index.php/login
 - You must be added to the instrument by the NanoBio lab manager after the equipment manager trains you.
- Equipment managers are listed on the 5th floor cork board in the common area. This is frequently updated. Always check first.
- Alternate managers are listed on the documentation for each piece of equipment.
- Plan your training! Make an appointment with lab staff & equipment managers in advance for training
 - o They will need to reserve time on their busy calendars and the equipment calendar.
 - Some training is best with real samples, ask your equipment manager.
 - o Take detailed notes on the operation, know the theory beforehand.
- Leave equipment clean and ready for the next person
- Report all equipment problems to the equipment managers and/or floor manager if there is no equipment manager.

D. Lab Jobs

• Everyone is responsible for keeping a clean, organized work space.

- Let Rita or the appropriate ordering person know when there is 1 pkg or rxn supplies. There are do
- Lab jobs should be done by the end of the week assigned or sooner if needed.
 - Dishes
 - ✓ Remove tape
 - ✓ Decontaminate and rinse *before* placing in the dirty dish basin.
 - ✓ Hand wash plastics that are incompatible dishwasher heat
- Equipment managers should check assigned equipment weekly.
- Lab meeting organizers should send out reminder emails to group the day before the meeting. As well as keeping the google schedule up to date.
- Media Makers should check the stocks and supplies weekly.
- MFe (Molecular Foundry E.coli strains) and MFm (Molecular Foundry microbes, non- E.coli) list
 organizers should keep lists up to date, check monthly, and ask group for additions.

E. Bench Courtesy and Lab Rules for Common Areas

- Keep yourself & your colleagues safe in the lab
 - o Emergencies call
 - √ 7-911 lab phone
 - √ 911 cell phone
 - ✓ Health Services ext. 6266
 - Eating, drinking, smoking, handling contact lenses, & applying cosmetics are not permitted in the lab.
 - Wash your hands with germacidal soap after handling viable materials, after removing gloves, and before leaving the laboratory.
 - No gloves should touch the door handles EVER.
 - o Work surfaces are decontaminated DAILY and immediately after a spill.
 - o The laboratory is designed so that it can be easily cleaned. So don't leave stuff around.
- Everyone is responsible for keeping a clean, organized work space.
 - o Promote this culture with your own actions!
 - Hey this was repeated it must be important.
- Any spills are cleaned up immediately.
 - o Chemical: Use appropriate clean up measures
 - o Biological: A bottle of disinfectant is kept in every work area for spills.
- If you do something wrong, confess.
 - This will establish you as an honest member of the community, which is an important quality in a scientific researcher!
 - o Offer to help with the remedy.
 - o Everyone makes mistakes.

- o No one gets mad at confessions!
- o This helps us fix problems more effectively than guess work!
- Never use reagents, buffers or pipettes without permission.
 - o They might be sterile or RNase free. They may be contaminated and ruin your experiment.
 - o If you are given permission to use a reagent, this is not a license to use whatever, whenever.
- If common reagent runs low **order more or let someone know** who orders that item (*before the bottle is empty*)
 - o This is especially important in a user facility.
- Don't ignore a broken piece of equipment, or an alarm.
 - o Notify the equipment manager, staff or your supervisor
- If someone is working unsafely, let them know!
 - Senior lab members too, or bad precedents will be set. If you don't feel comfortable, have your supervisor of floor staff do this for you.
 - o Safety especially important for students who are learning. **Be a good model.**
- Don't leave anything anywhere, except where it belongs on you lab bench
- Don't move common supplies
 - o This is user facility; people need to know where things are.
 - o If you have to move something (leave a note)
- Clean up immediately after each part of an experiment (or during if possible)
 - o Cleanup is part of the experiment, plan accordingly. (check these off)
 - ✓ Be especially careful of common areas.
 - ✓ Check daily waste containers after use.
 - ✓ Biosafety cabinets need to be wiped with 70% ethanol after use (and before).
 - ✓ Gel areas need to be wiped down. Gel boxes need to be rinsed.
 - ✓ Common reagents may need to be re-made.
 - ✓ Waste needs to be logged and stored properly.
 - ✓ Old cultures need to be decontaminated and dumped. Don't just leave them in the sink.
 - ✓ Don't let bio waste over fill the waste containers.
 - ✓ Remove your materials and samples from common areas and instruments and back to your bench
- Request a minimum of favors
 - o Ask *nicely* if you need someone do something.
 - o Be prepared if they say NO because of a conflict.

F. Laboratory Notebooks

Purpose of Notebook

The notebook's purpose is to record methods, observations, and results of your experiments so that you can go back and figure out what you did. It must be clear enough that another person can interpret your results. Therefore, a notebook must be clear and thorough

Ownership

Generally, the notebook should not leave the laboratory
The notebook belongs to the lab and institution

You are welcome to make digital or hard copies for your own records.

Type and Format of Notebook

- 1. The bound notebook used in our laboratory
- 2. Ink not pencil & Mistakes
 - a. Ballpoint pen with black ink is best
 - i. Uni-ball vision micro (waterproof, fade-proof)
 - b. Pencil writing can be erased (harder to prove authenticity)
 - c. Write legibly (you may also print out and paste in)
 - d. Mistakes?
 - i. Cross them out with a single line.
 - ii. White-Out and blacking out is not acceptable.
 - e. Paste all data in securely.

Content of the Laboratory Notebook

- 1. Table of Contents Save first three pages of notebook for Table of Contents. Include on each line
 - a. Title of Experiment
 - b. Date
 - c. Page Number
- 2. For each Experiment:
 - a. Date of Experiment
 - b. Title of Experiment
 - i. (e.g., Serial knock-down of DIAP1 by RNAi)
 - c. Purpose
 - i. (e.g., To determine the relationship between the amount of RNA- used against DNAP1 and the amount of cell death.)
 - ii. Articulate specific questions or goals in writing
 - d. Materials:
 - i. For new or the first time you are doing something, list the catalog # of specific chemicals or enzymes you are using
 - ii. Also, note any significant hazards of the chemicals, biologicals or processes you will be using.
 - e. Protocol
 - Write the protocol before you begin the procedure. Best practice is typed & pasted into your notebook.
 - ii. Amend with modifications, notes, and observations handwritten alongside.
 - iii. May refer to previous protocol in notebook (note any changes)
 - iv. Included any and all calculations
 - v. Important: Describe contents of all numbered tubes

- vi. Important:All tubes and data files should have systematic numbering which indicates the notebook page describing the experiment. Ajo-Franklin lab standard numbering: Name of notebook, page number, tube number (or data file number) → jc258001
- f. Observations and Results
 - i. Everything that happens or doesn't happen is data
 - ii. Any writing that will facilitate data entry should be planned out in advance
 - iii. May include: tables, charts, graphs, printouts, pictures, gels, films, calculations
 - iv. Important: All computer-based data must be centrally stored and systematically named
- g. Conclusion & Next Steps
 - i. Specifically, answer the questions or state progress towards goals.
 - ii. Prepare a conclusion. How did the experiment go?
 - iii. Next Steps: What were the unanswered questions? What are possible improvements?
- 3. Maintenance
 - a. Record everything ASAP
 - A. Weekly Check-Up
 - i. Attach data / printout / films
 - ii. Glue or double sided tape (not staples)
 - iii. Create tables and graphs
 - iv. Summary for the Week
 - v. Record experiment in the Table of Contents
 - vi. Make plan for the following week

G. Attendance and Vacation Policy

- a. Hours
 - i. You should strive to work the right number of hours to maximize your scientific productivity. If you work too few hours, you will have difficulty making sustained progress on your project; if you work too many hours, it will likely be counterproductive. In my experience, most people need to devote at least 40 hrs/week to make good research progress, but usually more than 80 hrs/week is counterproductive. (This includes seminar and meeting times.) You will need to find what works best for you.
 - ii. I expect everyone to be in lab during core hours: 10am-4pm Mon-Fri. Keeping shared hours allows more effective communication between lab members. Beyond these core hours, you have some flexibility in choosing what times you are most effective.
- b. Sick: If you are sick or will unexpectedly not be in lab, please also send Caroline and anyone else you will are scheduled to work with that day an e-mail letting them know you will not be in.
- c. Vacation: It is important to take time off. Please let Caroline know at least 1 week ahead of when you are planning to be gone for non-holiday periods. If you are an LBL employee (this includes summer interns (aka student assistants), postdocs, project scientists, and staff members), you can take vacation days as you earn them following the LBNL vacation policy for your position. If you are a UC Berkeley graduate student, you are allowed 1 month paid vacation (including holiday periods) and prior approval from Caroline is required.
- d. LBL Vacation Days:
 - i. Martin Luther King Holiday
 - ii. President's Day
 - iii. Cesar Chavez Day, Veteran's Day, or Dec 26
 - iv. Memorial Day
 - v. Independence Day
 - vi. Labor Day
 - vii. Thanksgiving Day and Day after Thanksgiving
 - viii. Christmas Eve Holiday
 - ix. Christmas Holiday
 - x. New Year's Eve Holiday
 - xi. LBL is closed from Dec 24-Jan 1, so besides listed holidays, vacation or leave without pay must be taken during this time

5. Supplies & Equipment

A. Ordering procedures:

- a. Some items are delegated to individuals for ordering, check the cork board in the common area first. If the item you need is assigned to someone for ordering, please email him/her and ask him/her to order more.
- b. Please check CMS and before ordering chemicals. https://cms.lbl.gov/jsp/login.jsp
 - i. It may be available internally.
- c. When should you ask before ordering?

- i. When the staff has specifically asked you to because budgets are tight.
- ii. When the item is more than \$200. More than \$300 requires authorization
- iii. If it it is a new chemical or material with a health, oxidizer, corrosive 3 or 4 hazardous material.

B. How to order:

- a. Use the electronic ordering system, ebuy: https://ebuy.lbl.gov
- b. If what you want is not on one of the ebuy vendor sites on try getting it added to ebuy first before doing a paper order.
- c. Under Government Scientific Source (GSS) click on "Add item to ebuy"
- d. You will get sent an email when it has been added (24-36 hrs)

6. Appendices

Appendix A: On-the-JobTraining (OJT)

Appendix B: Ajo-Franklin Biological Use Registration/Authorization (BUR/BUA)

Appendix C: Equipment Safety Data Sheets

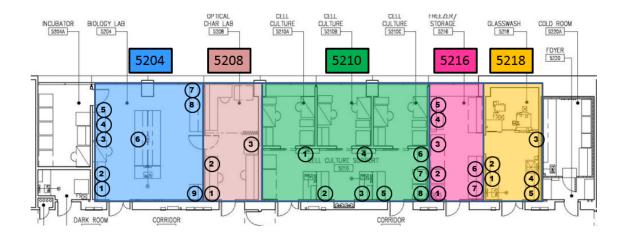
Appendix D: General Safety Resources

Appendix E: Commonly Used Hazardous Biochemicals

Appendix G: Directions to the Foundry

Appendix B: Bio-Side Supplies

Bio-Side Supplies



i. BioSide Supplies

Item	Location
PCR tubes, 0.2 mL	1 - 5204 (under bench)
Biohazard Autoclave Bags	2 - 5204 (under bench)
Nanowaste Bags	3 - 5204 (under bench)
Tube Racks	3 -5204 (Shelf Above bench)
Columns & Acessories	4 - 5204 (under bench)
Cuvettes (Disposable)	4 - 5204 (under bench)
NanoDrop Supplies	4 - 5204 (under bench)
Repeat Pipet Tips	4 - 5204 (under bench)
Agarose for DNA gels	5 -5204 (Shelf Above bench)
Repeat Pipet Tips	5 - 5204 (under bench)
Ziplock Bags	5 - 5204 (under bench)
Electroporation Cuvettes	6 - 5204 (under bench)
Electroporator	6 - 5204 (under bench)
Coulter Counter Tubes	6 - 5204 (under bench)
Timers	7 - 5204 (under bench)
Tape (Lab)	7 - 5204 (under bench)
Innoculating Loops	7 - 5204 (under bench)
Pipette Box (metal, autoclavable)	7 - 5204 (under bench)
Glass Culture Tubes	7 - 5204 (under bench)
Small Glass Beakers	7 - 5204 (under bench)

Ring Stand Clamps and Accessories	7 - 5204 (under bench)
VacuGuard Filters	7 - 5204 (under bench)
Strikers for Bunsen Burners	7 - 5204 (under bench)
Pipet Aide (old extra ones)	7 - 5204 (under bench)
Western Blot Apparatus and Accessories	8 - 5204 (under bench)
Diapers (Blue)	8 - 5204 (under bench)
Lab Bench Paper (Mat)	8 - 5204 (under bench)
DNA Gel Supplies	9 - 5204 (under bench)
Pipette Tips (Gel Loading)	9 - 5204 (under bench)
Vortex Accessories (Multi-tube Adaptors)	9 - 5204 (under bench)
Coulter Counter Tubes	9 - 5204 (under bench)
Racks	10 - 5204 (under bench)
Sonicator (Extra)	1 - 5208 (under bench)
Sealable Bottles and Stoppers	2 - 5208 (under bench)
Tubes (Glass Culture / Fraction Tubes)	3 - 5208 (Cabinet)
Syringes	3 - 5208 (Cabinet)
Needles	3 - 5208 (Cabinet)
Spin Filters / Columns	4 - 5208 (Shelf Above bench)
Gloves (Nitrile)	1 - 5210 (In shelf)
PBS Buffer	1 - 5210 (In shelf)
Seal Wrap	1 - 5210 (In shelf)
Tubes (50 ml Falcon) Polypropylene	
pH/DO, pH/Conductivity, pH/Ion Selective	1 - 5210 (In shelf)
Electode	2 - 5210 (under bench)
96 Well Assay Blocks	4 - 5210 (under bench)
96 Well 1.2 ml Tubes and Racks	4 - 5210 (under bench)
Shaker (Small Bench Top Size)	4 - 5210 (under bench)
Syringe Pump	4 - 5210 (under bench)
Plates (Multi-Well for Protein	
Crytsalization)	5 - 5210 (under bench)
Reagent Reservoirs (50 mL and 25 mL)	6 - 5204 (Shelf Above centrifuge)
Cell Culture Flasks	7 - 5210 (Cabinet)
Pipettes (5, 10, 25, 50ml and Aspirating)	7 - 5210 (Cabinet)
Filter Unit (1L, 0.22um)	7 - 5210 (Cabinet)
48 Well Plates, Non-Tissue Culture Treated	7 - 5210 (Cabinet)
Cryotubes	8 - 5210 (Cabinet)
Aluminum Foil	8 - 5210 (Cabinet)
Microfuge tubes, 1.5 mL clear	8 - 5210 (Cabinet)
Microfuge tubes, 1.5 mL colored	8 - 5210 (Cabinet)
Tubes (50 ml Falcon) Polypropylene	8 - 5210 (Cabinet)
Tubes (15 ml Falcon) Polypropylene	8 - 5210 (Cabinet)
Filter Unit (50, 250, 500 mL, 0.22um)	8 - 5210 (Cabinet)
Kimwipes	8 - 5210 (Cabinet)
Cling Wrap	8 - 5210 (Cabinet)

96 Well Plates Black	8 - 5210 (Cabinet)
Tubes (14ml Snap Cap Culture, Round	0 - 52 To (Odbinot)
Bottom)	8 - 5210 (Cabinet)
Glass Bottom Microwell dishes, 35 mm	9 F240 (Cabinat)
Petri, 10 mm Microwell	8 - 5210 (Cabinet)
Petri Dish, Falcon 35x10 mm Petri Dish, 60 mm, Sterile, Low-Cell-	8 - 5210 (Cabinet)
Binding	8 - 5210 (Cabinet)
Petri Dish, 100x15 mm, BD Falcon	8 - 5210 (Cabinet)
Tissue Culture Plates (Multi-Well)	8 - 5210 (Cabinet)
96 Well Plates, Clear, Polystyrene, Flat	o ozno (odbinot)
Bottom, Non-treated, Sterile	9 - 5210C (Shelf Above bench)
96 Well Plates, Clear, Flat Bottom, Non-	
Sterile, Bacti-Plate	9 - 5210C (Shelf Above bench)
96 Well Plate Sealers	9 - 5210C (under bench)
96 Well Plates, Black, Flat Bottom	9 - 5210C (under bench)
96 Well Plates, Black, Clear Bottom, Flat Bottom	9 - 5210C (under bench)
96 Well Plates, (Dilution Plates), "U"	o oznoc (under benen)
Bottom, Polyvinyl Chloride	9 - 5210C (under bench)
96 Well Plates, (Dilution Plates), Flat	0 F310C (under hench)
Bottom, Polyvinyl Chloride	9 - 5210C (under bench)
96 Well Plates, (Protein Crystallography) 96 Well Plates, "U" Bottom,	9 - 5210C (under bench)
Polypropylene	9 - 5210C (under bench)
96 Well Plates, Clear, "V" Bottom,	
Polypropylene	9 - 5210C (under bench)
96 Well Plates, Clear, Flat Bottom, Non- Sterile, EIA/RIA, Medium Binding	10 - 5210C (Shelf Above bench)
96 Well Plate Clear Plastic Lids	10 - 5210C (Shelf Above bench)
Plate Reader Manuals	10 - 5210C (under bench)
Tubes (5 ml Snap Cap), FACS Tubes,	10 - 52 100 (dilder belieff)
Polystyrene	10 - 5210C (under bench)
96 Well Plates, Black, "U" Bottom	10 - 5210C (under bench)
96 Well Plates, Black, Flat Bottom, Strip	40 F0400 (and an handa)
Wells	10 - 5210C (under bench)
Bio Nano Transportation Carriers	11 - 5210B (On bench)
Ice Machine	1 - 5216
Fermenter	2 - 5216
Solid Chemicals and Buffers	3 - 5216 (Cabinet)
Tubes and Accessories for Ultra Centrifuge	4 - 5216 (Shelf Above)
Pipetteman Tips (Standard Pipetman w/	
Barriers), 200, 1000 μL	5 - 5216 (Shelf Above)
Pipetteman Tips (Standard Pipetman w/o Barriers), 10, 20, 40, 200, 1000 μL	5 - 5216 (Shelf Above)
Pipetteman Tips (LTS Pipetman w/ &w/o	3 - 52 TO (Offeli Above)
Barriers)	6 - 5216 (Shelf Above)
Syringes	1 - 5218 (under bench)
Needles	1 - 5218 (under bench)
Surgical Masks	2 - 5218 (under bench)
Nalgene Bottles	2 - 5218 (under bench)
Naigerie Bottles	Z OZ TO (dildci bellott)

Updated 04 June 2013

Diaglia Daga	2 F240 (under hench)		
Plastic Bags	2 - 5218 (under bench)		
Aluminum Block	2 - 5218 (under bench)		
Heat Block	2 - 5218 (under bench)		
Langmuir Trough Supplies, 50 mL			
Polypropylene Tubes	3 - 5218 (under bench)		
Culture Tubes	4 - 5218 (under bench)		
pH Meter	4 - 5218 (under bench)		
Gradient Maker & Fractionator, Parts,			
Manual	5 - 5218 (under bench)		
Balance	5 - 5218 (under bench)		
		3	-
			5220
Qiagen Kits (Miniprep, PCR, Gel			(Cab
Extraction)			inet)

Appendix C: Locations of Commonly Used Biochemicals

Chemical manage ment system is useful for seeing if we have it: https://cms.lbl.gov/jsp/login.jsp

a. Dry Chemicals: 5201, 5216, 5218

b. Detergents: 5208c. Media: 5218

d. Acids / Bases: 5216, 5201

e. Solvents, including ethanol and acetone: 5201

Appendix D: Equipment Safety Data Sheets

NanoBio Equipment Safety Information Sheet

Updated ____

Equipment:	
Equipment Manager:	
Manager Phone:	
Manager e-mail:	
Back-up Contact/Phone:	
PART 1. Safety and Controls	
Personal Protection Equipment	
2. Waste Disposal Notes:	
Chemical Hazards:	
4. Biological Hazards:	
4. Biological Hazards.	
5. Laser Hazards:	
6. Electrical Hazards:	
7. Mechanical Hazards:	
8. Engineered Nanomaterials:	
Other Safety Guidelines:	

PART 2. General Directions and Important Notes							
•	•						
•							
•							
•							
•							
PART 3: List of Authorized People	e						
You may not use this instrument unless your name is listed below. Please direct requests for authorization and questions to the instrument manager.							

Appendix D: General Safety Resources

Useful links to LBL and other sources on Biosafety, Chemical Hygiene, and Nanomaterials

ES&H MANUAL AKA LBNL PUB 3000

http://www.lbl.gov/ehs/pub3000/

Chemical Hygiene Plan

http://www.lbl.gov/ehs/chsp/index.shtml

Biosafety:

http://www.lbl.gov/ehs/pub3000/CH26.html#2651 http://www.lbl.gov/ehs/biosafety/manual/index.shtml

Biosafety in Microbiological and Biomedical Laboratories (BMBL) 5th Edition (CDC)

The NIH-CDC standard <u>Biosafety in Microbiological and Biomedical Laboratories</u> (**BMBL**) is the national code of practice and Berkeley Lab biosafety standard that outlines and defines biosafety risk assessment and control. http://www.cdc.gov/biosafety/publications/bmbl5/index.htm

Nanomaterials:

http://www.osha.gov/Publications/OSHA_FS-3634.pdf http://www.lbl.gov/ehs/chsp/html/nanomaterials.shtml

BIOHAZARD LABORATORY CHECKLIST Lawrence Berkeley National Lab EH&S Industrial Hygiene - Biosafety

Principal Investigator:	Biosa	fetv	Reg. #:				
Division: Labs/Rms. Inspected:			. 109. 11.				
Inspected D							
Based on an inspection this day, items indicated below identify viol	Date:	,					
Televine and a report to the second state of the second se	<u>iideline</u>	e for E	Pesarch involving				
Recombinant DNA Molecules" (2001).	auchite.	3 /U/ I	research Mildoldling				
ALL BIOSAFETY LEVELS							
COMPLIES	YES	NO	COMMENTS				
Standard Microbiologic Practices and			COMMENTS				
1. Lab personnel understand biology of organisms used in the lab and	1	T -					
nave received training in aseptic technique.		1	1				
2. The lab has an emergency action plan that describes procedures in	1.						
the event of an accident and the lab personnel are familiar with it.							
3. Access to lab is limited or restricted at the discretion of the lab Pl	-						
he lab bears a biohazard label indicating the agent(s) and Rel		٠,					
4. Work surfaces are decontaminated once a day and after any shill of							
viable material.							
5. All contaminated liquid is decontaminated before disposal. Solid							
medical waste is handled according to the BSL (autoclaved onsite or sent out for treatment)							
6. Mechanical pipetting devices are used; mouth pipetting is prohibited.							
7. Eating, drinking, smoking, and applying cosmetics are not permitted in	1						
the work areas. Food may be stored in cabinets or refrigerators							
designated and used for this purpose only. Food storage cabinets should be located outside of the work area.							
8. Persons wash their hands after they handle viable materials or							
animals (after removing gloves) and before leaving the laboratory.							
Policies for safe handling of sharps are instituted.							
10. All procedures are performed carefully to minimize the creation of	 	ļ					
aerosols.							
Laboratory Facilities		<u> </u>					
11. The lab is designed so that it can be easily cleaned	,						
 Bench tops are impervious to water and resistant to the chemicals 	 	<u> </u>					
used in the area. Lab furniture is sturdy.			İ				
3. Spaces between equipment are accessible for cleaning & are clean	:						
14. Each lab contains a sink for hand washing. Hand soan is at the sink	 						
5. Openable windows are fitted with fly screens.							
BIOSAFETY LEVEL 1 (BSL-1	100	\					
COMPLIES COMPLIES							
	YES	NO					
6. Lab gowns or coats are recommended.	١						
7. Contaminated materials that are decontaminated at a site away from							
the lab are placed in a red biohazard bag within a durable leak-proof							
container that is closed before removing from the laboratory.							
8. Personnel know of the requirement for an insect and rodent control	ļ						
program. [If pest control is needed, call Facilities' Work Request @ 6274.]							
Tork Toquest @ 0274.]	<u> </u>						

Appendix E: Materials Safety Data Sheets (MSDS) of commonly used Hazardous Biochemicals:

By all means this is *not* a complete list. Please remember that being prepared for lab includes knowing the potential hazards of the chemicals, biologicals or processes you will be using. This includes the waste disposal. For example, if you have just saturated a biological solution with ammonium carbonate you don't want to kill the microbe with bleach. The ammonia and sodium hypochlorite will react to form chloramine and other compounds which are very irritating and reactive toxic gases.

NH₃ + NaOCl → NaOH + NH₂Cl

Here are a few <u>from http://bitesizebio.com/articles/ten-bad-chemicals-in-the-lab-and-what-they-do-to-you/</u> accessed 05/08/13, and some others specific to our lab.

1. Acetonitrile

Be careful with this flammable irritant. Once this solvent is inhaled, ingested, or absorbed through the skin, it converts to cyanide!

National Center for Biotechnology Information. Acetonitrile – Compound Summary. PubChem Compound. Available at: http://pubchem.ncbi.nlm.nih.gov/summary/summary.cgi?cid=6342&loc=ec rcs#x332 Sigma-Aldrich. Material Safety Data Sheet: Acetonitrile. 2012. Available at: http://www.sigmaaldrich.com/catalog/product/sial/271004?lang=en®ion=US

Salting-out assisted liquid/liquid extraction with acetonitrile: a new high throughput sample preparation technique for good laboratory practice bioanalysis using liquid chromatography-mass spectrometry. *Biomed Chromatogr.* 2009;23(4):419-425.

2. Chloroform

This volatile solvent can irritate the skin, eyes, and lungs. It also acts as an anesthetic that depresses the central nervous system. Once inside the body, it converts to highly toxic phosgene, a chemical weapon used during World War I.

Luttrell WE. Toxic tips: chloroform. Chem Health Safety. 2005;12(3):36-37.

3. Dimethyl Sulfoxide (DMSO)

DMSO is such an excellent solvent that it crosses healthy, intact skin – and takes whatever is dissolved along with it! Be sure to wear your butyl rubber gloves if you are dissolving large amounts of something toxic (such as the neurotoxic pesticide rotenone) in DMSO. Sigma-Aldrich. Material Safety Data Sheet: Dimethyl Sulfoxide. 2012. Available at: http://www.sigmaaldrich.com/catalog/product/sigma/d8418?lang=en®ion=US

4. Formaldehyde

This common fixative is a suspected human carcinogen. Take advantage of the fume hood, because formaldehyde can cause dermatitis, sinusitis, and asthma! And don't buffer formaldehyde with hydrochloric acid, because together they form a potent carcinogen, bis-chloromethyl ether.

5. 2-Mercaptoethanol

As if the rotten-fish smell wasn't bad enough, 2-mercaptoethanol is a combustible corrosive. It can harm the skin and the mucous membranes, and cause larynx spasms, pneumonitis, and pulmonary edema when inhaled. Sigma-Aldrich. Material Safety Data Sheet: 2-Mercaptoethanol. 2012. Available at:http://www.sigmaaldrich.com/catalog/product/sigma/m3148?lang=en®ion=US

6. Methanol

Like other volatile solvents, methanol can easily enter the body through the lungs, gut, or skin. Once inside, methanol transforms to formic acid, which causes metabolic acidosis and blinding retinal toxicity.

7. Sodium Azide

This popular preservative is an extremely toxic skin irritant that can cause headaches, dangerously low blood pressure, and even heart failure. Sadly, its toxicity and ready availability in labs have made it a method of suicide for researchers (http://www.ncbi.nlm.nih.gov/pubmed/22559996). Another word of caution: don't pour sodium azide down the sink where it can react with copper and lead pipes, forming highly explosive substances! Human health effects of sodium azide exposure: a literature review and analysis. *Int J Toxicol*. 2003;22:175-186.

8. Sodium Cholate

You might think, ah this is just a detergent and it tis rate as 0,0,0 for health, fire and reactivity. However, because it tis sold as a powder and is used to dissolve membranes it is really irritating to respiratory tract membranes in you! Avoid dust!

MSDS:

http://www.sigmaaldrich.com/catalog/search?interface=All&term=sodium+cholate&lang=en®ion=US&focus=product&N=0+220003048+219853269+219853286&mode=match%20partialmax

9. Sodium Hydroxide

Disturbingly, it's better to be splashed in the eye with concentrated acid than sodium hydroxide. Acids precipitate proteins, which form a protective "scab" over unharmed tissue, but strong bases like sodium hydroxide saponify fatty acids and destroy cell membranes. The "scab" never forms, so the base can just keep burning its way through. Wear your goggles!

Agency for Toxic Substances and Disease Registry. ToxFAQs[™] for Sodium Hydroxide. Toxic Substances Portal. 2011. Available at: http://www.atsdr.cdc.gov/toxfaqs/tf.asp?id=248&tid=45
Chang S, Lamm SH. Clark DE. Chemical injury to the eye. *Chem Health Safety*. 2002;9(2):6-9. National Institute for Occupational Safety and Health Education and Information Division. Sodium hydroxide. NIOSH Pocket Guide to Chemical Hazards. 2011. Available at:http://www.cdc.gov/niosh/npg/npgd0565.html

10. Sodium Hypochlorite

In solution, this becomes bleach – an excellent anti-microbial because it is a strong and corrosive oxidant. While most researchers have gotten a burning whiff (and maybe a splash) of this irritant, they may not know that bleach can actually cause allergic contact dermatitis. Future exposure can then trigger skin reactions to even dilute bleach.

Luttrell WE. Toxic tips: sodium hypochlorite. *Chem Health Safety*. 2001;8(6):24-26.

11. Tetrahydrofuran (THF)

THF is a flammable solvent. Over time, THF produces shock-sensitive, explosive peroxides. If the THF evaporates off, the peroxides will concentrate in the remaining solution. Even slight bumping of a container containing concentrated peroxides can result in an explosion.

(http://www.ehs.uci.edu/salerts/Lesson%20Learned Peroxide.pdf).

Sigma-Aldrich. Peroxide Forming Solvents. Sigma-Aldrich Learning Center. 2013. Available at: http://www.sigmaaldrich.com/chemistry/solvents/learning-center/peroxide-formation.html

Zhang J, Wu H, Kim E, El-Shourbagy TA.

Appendix F: Sexual Harassment Policy

The Laboratory is committed to creating and maintaining a community in which all persons who participate in Laboratory programs and activities can work together in an atmosphere free from all forms of harassment, exploitation, or intimidation, including sexual. Specifically, every member of the Laboratory community should be aware that the Laboratory is strongly opposed to sexual harassment and that such behavior is prohibited by law and by Laboratory policy. It is the intention of the Laboratory to take whatever action may be needed to prevent, correct, and, if necessary, discipline behavior that violates this policy.

Link to full policy below:

http://www.lbl.gov/Workplace/RPM/R2.01.html#secb

Appendix G: Transportation, Directions to the Foundry & Map of LBL Site

Directions to Foundry from campus:

The best way to get here is to take the Orange Route of the LBL Shuttle bus to to Bldg. 62. There are a variety of shuttle stops (see the map below), perhaps the easiest to access is the East Gate bus stop. You will need to show the driver your UC Berkeley ID and tell him/her that you're coming to Bldg 67 to board the bus. The Foundry (Bldg. 67) is just a few minutes walk from the bus stop at 62. Ask the bus driver to point you in the right direction. And if you run into any problems, feel free to give me a ring in my office or on my cell (#s are below). A map of the shuttle bus stops and routes can be found here:

http://www.lbl.gov/Workplace/Facilities/Support/Busses/all routes.html.

Directions to LBL via BART & bus:

From the Downtown Berkeley BART station, go to the LBNL bus stop at the corner of Shattuck and Addison. Then take the Orange LBNL Shuttle

to Bldg. 62 (you will need to show them the gate pass). The Foundry (Bldg. 67) is just a few minutes walk from the bus stop at 62. Ask the bus driver to point you in the right direction.

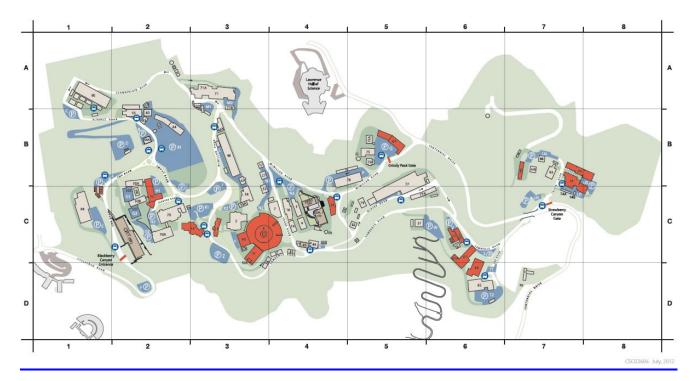
A map of the shuttle bus stops and routes can be found here:

http://www.lbl.gov/Workplace/Facilities/Support/Busses/all routes.html

The location of the LBL entrance nearest to the Foundry, the Strawberry Canyon Gate can by mapping directions to Strawberry Canyon Gate, Berkeley, CA.

When you get to Strawberry Gate just pull over to the right and give them your name and they will issue you a parking pass. Turn left once you enter the gate and park along the main road wherever you can. The Foundry entrance (C6 on map below) is located along the main road; our labs and offices are located on that entrance floor (the 5th).

http://www.lbl.gov/Workplace/lab-site-map.html



LBL Shuttle

Next Bus Online predictor

http://www.nextbus.com/predictor/stopSelector.jsp?a=lbl

Route Maps

